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The Incidence and Intensity of Haematozoa in the Common Crow (Corvus brachyrhynchos) of Western Washington

Ronald E. Scarvie
Central Washington University

THE INCIDENCE AND INTENSITY OF HAEMATOZOA IN THE COMMON CROW (*CORVUS BRACHYRHYNCHOS*) OF WESTERN WASHINGTON

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by

Ronald E. Scarvie

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APPROVED FOR THE GRADUATE FACULTY

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Glen W. Clark, COMMITTEE CHAIRMAN

_____

Dan A. Unruh

_____

Curt A. Wiberg

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Chapter 1

INTRODUCTION

The purpose of this study was to determine the incidence and intensity of blood parasites in the common crow (Corvus brachyrhynchos). The following observations on the protozoan genera Haemoproteus, Leucocytozoon, Plasmodium and Trypanosoma with observations on the nematode group microfilaria, are based entirely on materials collected at Auburn, Washington during the spring of 1970.

Despite the numerous studies on prevalence of blood parasites in various birds, there have been few attempts to study the relative incidence and intensity of haematozoa in blood and tissue smears taken from the common crow. So far as the author has been able to ascertain, no studies of this nature have been recorded previously from Western Washington.

Chapter 2

REVIEW OF THE LITERATURE

Blood protozoa were first reported from avian hosts by Danilewsky in 1885 from the Russian Ukraine, but no investigations were made in North America until the summer of 1897 when Opie (27) and MacCallum (19) studied blood smears of birds from Maryland and Ottawa. Novy and MacNeal (25) published in 1904 and 1905 on blood parasites of Michigan birds. In 1913 Whitmore (33) isolated a strain of Plasmodium from "sparrows" in New York. The Department of Protozoology and Medical Entomology of the Johns Hopkins University School of Hygiene and Public Health began an extensive study on the biology of avian Plasmodium in the 1920's (13). In recent years related investigations have been undertaken on other blood parasites of birds. The first extensive survey in North America was made on ducks in Michigan by O'Roke (28). The first extensive survey of a variety of wild birds was conducted in the region around Syracuse, New York (22, 23). The most extensive survey of avian haematozoa in Central Washington was undertaken by Clark, Lee and Lieb in 1968 (6).

The haematozoa Haemoproteus, Leucocytozoon, and Plasmodium belong to the subphylum Sporozoa with Haemoproteus and Leucocytozoon belonging to the family Haemoproteidae, Plasmodium to the family Plasmodiidae.

Trypanosoma avium, a hemoflagellate, belongs to the protozoan class Zoomastigophorea. Microfilaria are embryos of filaria worms belonging to the superfamily Filarioidea of the sub-order Spirurata.

The genus Leucocytozoon is found only in birds. Leucocytozoon gametocytes live in white blood cells whose nuclei are pushed aside and flattened shortly after parasite penetration (4). The only known vectors for Leucocytozoon belong to the genera Simulium and Culicoides (4). There have been few reported infections of Leucocytozoons in the common crow (6, 8, 11, 1, 32). The genus Haemoproteus occurs in birds and reptiles. It parasitizes the red blood cell. The elongated, halter-shaped gametocyte curves around the nucleus of the red blood cell (3). The vectors of Haemoproteus are probably pupiparous flies of the family Hippoboscidae and flies of the genus Culicoides (3). The incidence of Haemoproteus is relatively high in the common crow (11, 32). Love, Wilkens and Goodwin in a study made in Georgia in 1953, found 5 crows infected with Haemoproteus of 15 examined. In 1960, Bennett and Fallis examined 10 immature crows and found 5 infected. Morgan and Waller (24) in 1941, and Jones (16) in 1968, found a very low incidence of both Leucocytozoon and Haemoproteus infection in the common crow. Jones (16) found a higher percentage of protozoan infection in immature crows than adult crows.

The blood protozoan Plasmodium is most readily observed within the red blood cell. "The bird parasites of the genus Plasmodium are of particular interest because of their extensive use in the study of the biology and treatment

of malarial parasites in general. It was in P. gallinaceum of chickens that exo-erythrocytic stages were found to be an essential part of the life cycle of malaria parasites" (3). The gametocytes of Plasmodium are found in the red blood cells; schizogony occurs in erythrocytes and the exo-erythrocytic forms are found in various tissues (3). The vectors of Plasmodium in birds are Culicine or Anopheles mosquitoes (13). The only record of Plasmodium infection in the literature for the common crow was reported by Love, Wilkens and Goodwin in 1953 (18).

Lavern, in 1903, described a trypanosome which he found in an owl (10). He classified the organism as Trypanosoma avium. The species name has since been ascribed to a number of trypanosomes found in a variety of birds. These flagellated organisms are extracellular and in birds have been recorded in the blood and bone marrow (30). In general morphology they are similar to the trypanosomes which occur in other vertebrates. In birds, these organisms are known to be extremely pleomorphic and, therefore, are difficult to differentiate on the basis of morphology alone (13). The vectors of T. avium are probably mosquitoes and hippoboscids (26). Stabler, Holt and Kitzmiller (30) examined four crows from Colorado in 1966, and found 2 infected with T. avium. Other investigators have found T. avium in the common crow from small samplings (6, 11, 1). Microfilaria are colorless, transparent embryos of filarial worms that may or may not be inclosed in sheaths (3). This name is given to microfilaria of various filariids of which the adult stage is not known (17). Microfilaria have been observed in several species of birds (5, 6, 7, 20, 21, 31).

Wetmore (32) in 1940, examined 16 crows collected in Washington D. C. and found 4 infected with Microfilaria. In 1953, Love, Wilkens, and Goodwin (18) examined 15 crows from Georgia and found 4 infected, and Clarke (8) in 1946, examined 4 crows and found 1 infected with microfilaria. Robinson, in 1954, reported that "the average maximum for crow infection by microfilaria is 80% but this will fall to about 40% during the breeding season" (29). Boughton, Bird and Lund found that "Dilution counts on samples of day and night peripheral blood show a marked nocturnal periodicity in the natural microfilarial infection in crows" (2).

Chapter 3

MATERIALS AND METHODS

Blood and tissue smears were taken from adult common crows (Corvus brachyrhynchos) collected in the vicinity of Lake Holm three miles southeast of Auburn, Washington. Adult crows were shot and as soon as possible after death thin blood films were made from one of several peripheral areas and from the heart. Tissue smears were also taken from small sections of the lung. Prior to making the smear, each piece of tissue was wiped clean of blood, macerated on the slide and then all but a thin smear was removed. Blood and tissue smears were air dried, fixed in absolute methyl alcohol and stained with Giemsa. The marrow from the 63 crows was checked in buffered water for living trypanosomes. Most marrow was examined within 1-3 hours after the death of the host. All marrow samples were taken from the femur.

All stained smears were examined for at least 15 minutes: 5 minutes with low power (200x), 5 minutes with medium power (430x) and 5 minutes with the oil immersion lens (1000x). These three magnifications greatly increased the probability of finding haematozoa in both blood and tissue preparations. Photomicrographs were taken on 35mm Panatomic-X film. All measurements were made with an ocular micrometer.

Chapter 4

RESULTS

A summary of the results of this and a number of previous studies appears in Table 1.

A total of 63 crows were examined. All 63 birds were parasitized with at least one species of blood parasite. The parasites identified were Leucocytozoon, Haemoproteus, Plasmodium, Trypanosoma and microfilaria. Multiple infections involving two or more blood parasites were common. Of a total of 63 birds, 8 carried a single parasite, 38 carried 2 species, 15 carried 3 species, and 2 harbored 4 species. Leucocytozoon and Haemoproteus occurred in the same bird 54 times, Leucocytozoon and Plasmodium occurred together 13 times, and Leucocytozoon, Trypanosoma and microfilaria were found in the same bird 4 times.

Leucocytozoon (Figures 1, 2) was found in 63 (100%) of the birds examined. The host cell nucleus, which stained a deep reddish purple, was deformed and pushed to the outside of the cell, forming a band along one-third to two-thirds of the periphery of the parasite. In some cases the host nucleus had been divided and appeared as crescents at opposite sides of the parasite. The intensity of the infections were light to heavy (3-111 organisms per 15 minute search period). Twenty-five organisms were measured. The length varied from 11.0 to 16.5u, with an average of

Table 1

Summary of Blood Parasites Reported for the Common Crow

Authority	Area	Examined	Total Positive	Plasm.	Leuc.	Tryp.	Mf.	Hp.
Stabler, Holt								
Kitzmiller 1966	Colorado	4	2			2		
Wetmore 1940	Wash. D. C.	16	7		1		4	2
Clark, Lee, Lieb 1968	Washington	2	1		1	1		
Coatney, West 1938	Nebraska	1	1		1	1		1
Coatney, Jellison 1940	Montana	1	1					1
Herms, Kadner, Galindo, Armstrong 1939	California	2	0					
Hart 1949	South Carolina	1	1					1
Manwell, Herman 1935	New York	2	0					
Love, Wilken, Goodwin 1953	Georgia	15	5	1			4	5
Clarke 1946	Toronto	4	3		3		1	
Bennett, Fallis 1960	Ontario	10*	8		8	2		5
Herman 1938	Cape Cod	4	2					2
Morgan, Waller 1941	Iowa	112	7	1	3	1	1	1
Jones 1968	Ohio	339	13		12			1
Scarvie 1970	Washington	63	63	13	63	4	4	54

*Immature birds.

Abbreviations: Plasm. = Plasmodium; Leuc. = Leucocytozoon; Tryp. = Trypanosoma; Mf. = Microfilaria; and Hp. = Haemoproteus.

Figure 1. Leucocytozoon macrogametocytes showing displacement of host-cell nucleus. All blood smears were stained with Giemsa. (1000x) HN = host nucleus, P = Parasite.

Figure 2. Leucocytozoon microgametocyte. Legend is the same as for above figure.

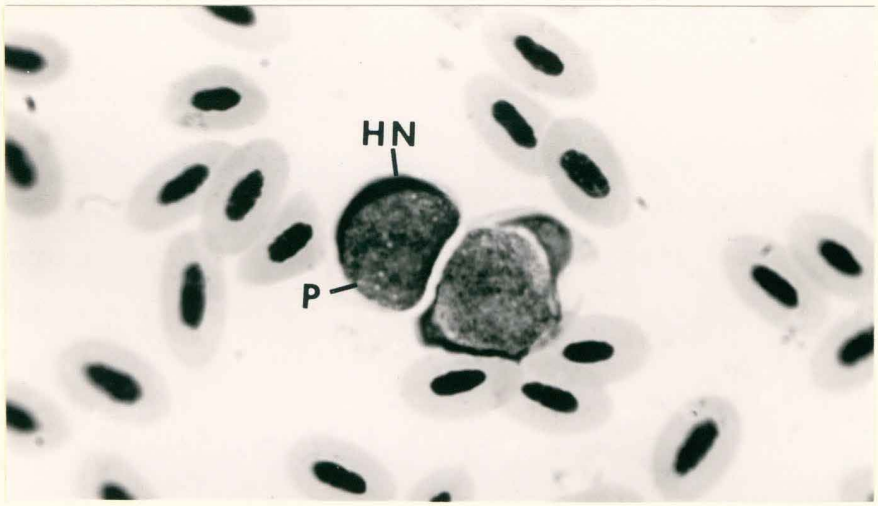

Figure 1

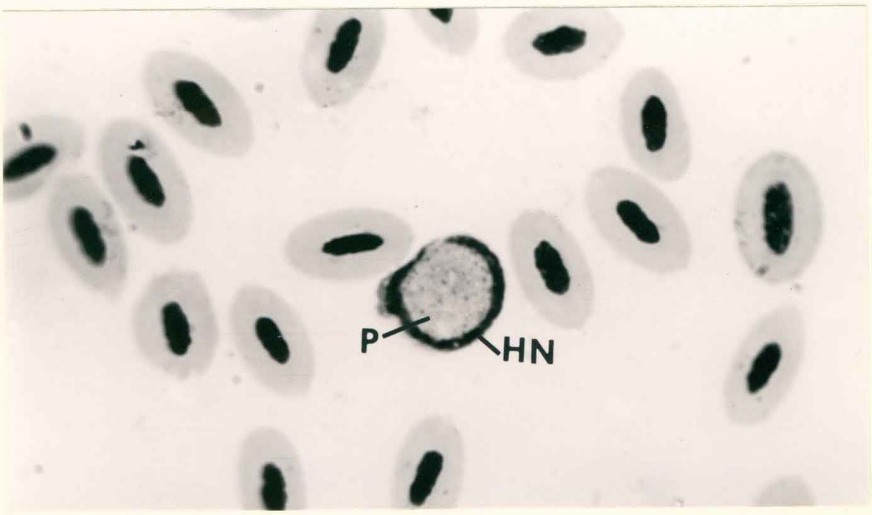

Figure 2

12.9u. The width varied from 5.5 to 15.4u with an average of 10.6u. The size of the nucleus ranged from 1.1 to 4.4u, with an average of 2.7u.

Haemoproteus (Figures 3, 4) was found in 54 (86%) of the birds examined. The parasite grows on one side of the host cell forming a horse-shoe shape around one side of the host cell nucleus. The nucleus was sometimes displaced very slightly. The intensity of each infection was light. Twenty-five organisms were measured with the length varying from 11.0 to 15.4u, with an average of 12.2u. The width ranged from 2.2 to 6.6u, with an average of 2.6u. The size of the nucleus varied from 1.1 to 3.3u, with an average of 1.2u.

Plasmodium was found in 13 (21%) of the birds examined. The intensity of each infection was very light.

Trypanosoma (Figure 5) was found in 4 (6.5%) of the birds examined. The intensity of each infection was very light. No Trypanosoma were found in the bone marrow preparations. Eight organisms were measured with the length varying from 37.4 to 45.1u, with an average of 43.6u. Width at the nucleus varied from 3.3 to 7.7u, with an average of 5.6u. The distance from the Karyosome to the posterior end ranged from 7.7 to 17.6u, with an average of 14.2u. The size of the nucleus varied from 2.2 to 5.5u in diameter with an average of 4.9u.

Microfilaria (Figure 6) were found in 4 (6.5%) of the birds examined. The intensity of infection was very heavy in one bird (Figure 7) and light in the other 3. Twenty-five organisms were measured. The length varied from 127.0 to 174.2u, with an average of 147.5u. The width ranged from 3.9 to 6.5u with an average of 5.1u.

Figure 3. Haemoproteus macrogametocyte. HN = host-cell nucleus, P = parasite (1000x).

Figure 4. Haemoproteus microgametocyte. Legend is the same as for the above figure. (1000x).

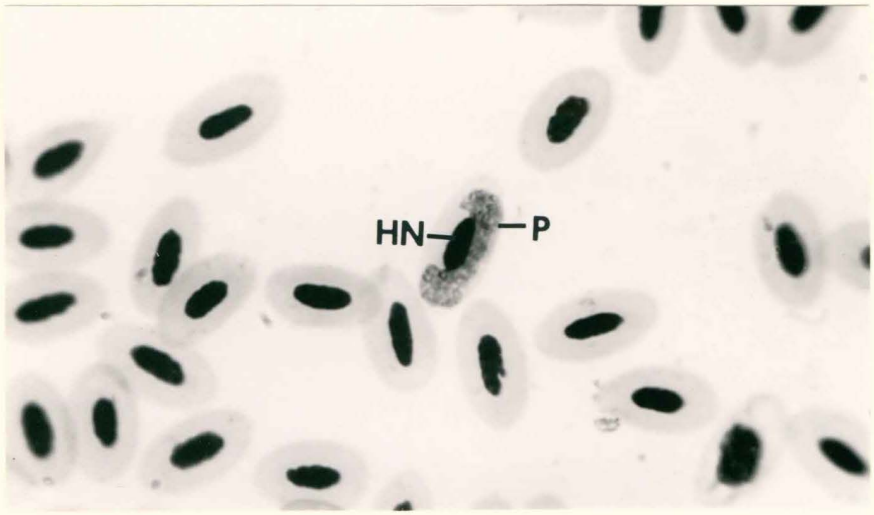

Figure 3

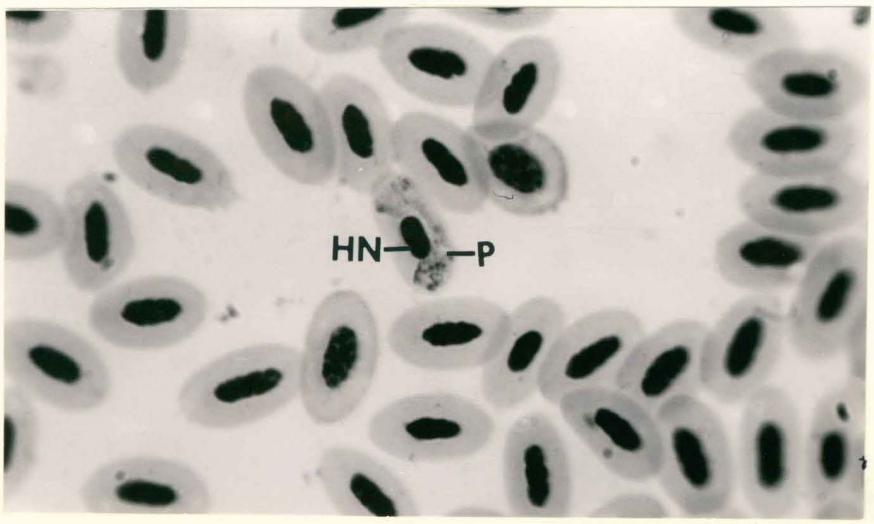

Figure 4

Figure 5. Trypanosoma sp. from peripheral blood of the crow. K = karyosome, N = nucleus. (1000x).

Figure 6. Microfilaria from peripheral blood of the crow. M = microfilaria. (400x).

Figure 7. Photograph showing a heavy infection with microfilaria. M = microfilaria. (100x).

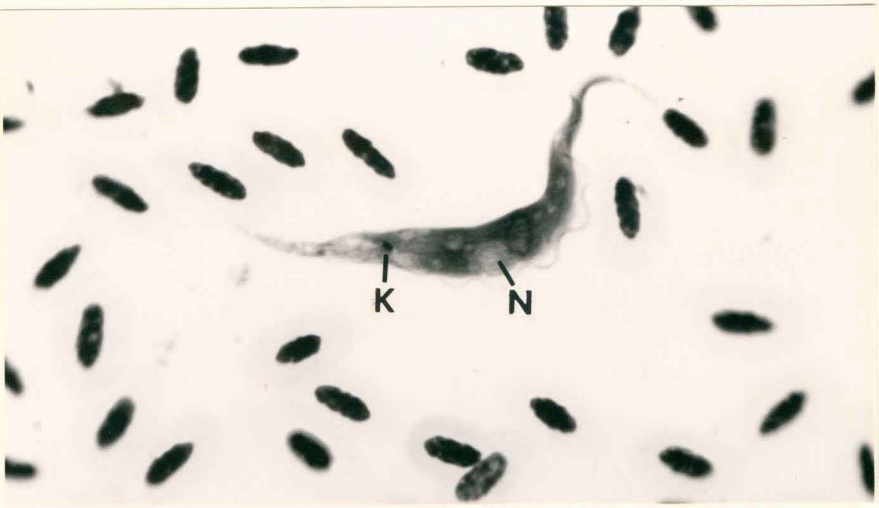

Figure 5

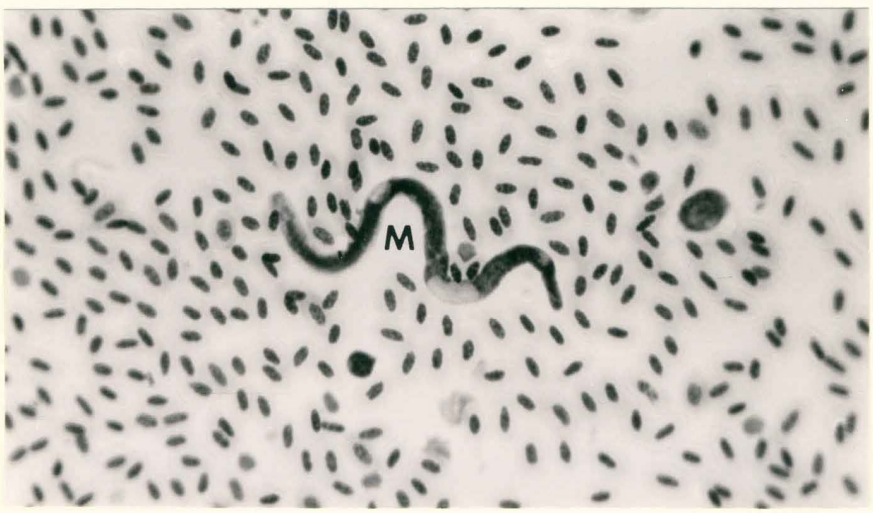

Figure 6

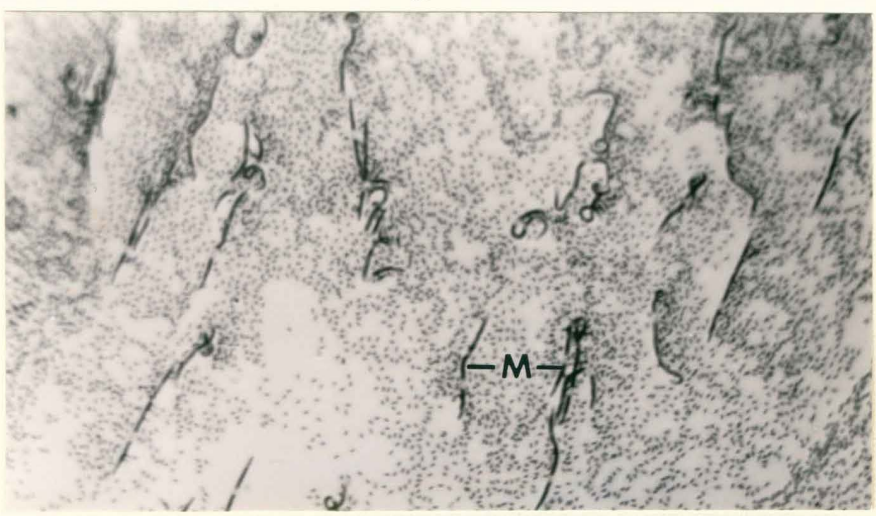

Figure 7

A higher incidence of Leucocytozoon infection was found in lung smears than in peripheral or heart smears. The highest incidence of Haemoproteus was found in blood smears taken from the heart. Plasmodium incidence was highest in peripheral and heart smears. The highest incidence of microfilaria was found in the lung smears. The incidence of Trypanosoma was very low in both blood and tissue smears.

Chapter 5

DISCUSSION

The high incidence of Leucocytozoon and Haemoproteus infection found in the common crow agrees with findings by other authors working with blood parasites of other species of birds (1, 4, 20, 34). It is noteworthy however that Leucocytozoon was detected in all crows examined, and Haemoproteus was found in 85% of the crows examined. A 100% Leucocytozoon infection has never been reported in the literature for any species of birds (providing a large sample was taken) (5, 20, 21, 4, 7), and it is especially an uncommon occurrence in crows (9, 12, 18). Morgan and Waller, in 1941 (24), examined 112 crows and found 3 infected with Leucocytozoon and 1 with Haemoproteus. In 1968, Jones (16) examined 339 crows and found 12 positive for Leucocytozoon and 1 positive for Haemoproteus. The findings of Morgan and Waller and Jones do not concur with the findings of the author.

The low incidence of Trypanosoma in the blood of the common crow concurs with the findings of other authors for different species of birds (5, 7). The fact that no T. avium were observed in the bone marrow preparations might well be explained on the basis that bone marrow preparations were examined without the aid of a phase microscope. It is quite possible that the author failed to observe the presence of T. avium because of inadequate equipment.

The relatively low incidence of Plasmodium in the common crow (21%) would agree with the current findings by other authors on different species of birds. Huff (15), who examined 278 birds, belonging to 13 species of Fringillidae (chiefly from Illinois) found only six cases in the lot. Herman et al. (14) examined 530 finches from the southwestern United States and found 51 positive for Plasmodium. Clark (5) examined 156 yellow-billed magpies from the vicinity of Davis and Gustine, California, and found 34 infected with Plasmodium. Plasmodium has been observed only one other time in the common crow (18). The low incidence in some reported studies could be due to the small sampling of crows examined.

Microfilaria are common parasites of many species of birds but they are not always seen in blood films since many of them are nocturnal and are not found in peripheral blood during the day (2). The present findings indicated a higher incidence in contact preparation from lung tissue than in peripheral blood smears. This agrees with the findings of Manwell (20) and Clark (5). The intensity of infection with microfilaria was also greater in lung smears than in blood. The relatively low incidence of microfilaria detected in the crow (6.5%) agrees with the findings of other researchers (1, 6, 9).

The data compiled by the author raises two questions of importance: 1) Will a change in season cause a difference in incidence and intensity of Haematozoa in the common crow? and 2) Would an examination of immature crows of western Washington reveal the same incidence and intensity of blood parasites as did Jones' (16) study of Haematozoa of Ohio?

Chapter 6

SUMMARY

1. A total of 63 crows were examined. All 63 birds were parasitized with at least one species of blood parasite. The parasites identified were Leucocytozoon, Haemoproteus, Plasmodium, Trypanosoma and microfilaria.

2. Multiple infection involving two or more blood parasites were common. Of a total of 63 birds, 8 carried a single parasite, 38 carried 2 species, 15 carried 3 species and 2 harbored 4 species.

3. Leucocytozoon was found in 63 (100%) of the birds examined, Haemoproteus in 54 (86%), Plasmodium in 13 (21%), Trypanosoma in 4 (6.5%) and microfilaria in 4 (6.5%).

4. Trypanosoma was not found in the bone marrow preparations.

5. A higher incidence of Leucocytozoon and microfilaria infection was found in lung smears than in peripheral or heart smears.

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